

institut für informatik



Heart Analysis Tool

User Manual

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1 Getting Started

This user manual gives you a detailed overview of the *Heart Analysis Tool*. It provides you with all necessary information how to use the program and it gives you a short introduction to the algorithms which have been used.

To start the program simply double click on the executable *Heart Analysis Tool.exe*. No further files or configurations are required. If the program does not start or if it throws any error messages, please have a look at the System Requirements (2) or Trouble Shooting (6).

As the application needs no additional files and only 3 MB of disk space, it can be sent via mail or it can be put on an USB device. It can be transferred to other systems, it does not need any installation and it can be even started from a network drive without problems.

In the following chapters keywords like methods, operations and buttons are printed in capital letters. In the majority of cases the reference to the specific method is placed in brackets behind the keyword.

2 System Requirements

Due to time pressure it was not possible to test the program on all systems but it should work well with a video card with at least 64 MB of memory. Especially when using a notebook or any other systems with a video card which uses shared memory you should read the Trouble Shooting (6). A fast CPU is advantageous.

2.1 Minimal Requirements

OS: Windows XP (SP1)

CPU: 1.5 GHz

RAM: 512 MB

Hard Drive: 5.0 MB or more free space

Video: Video Card with 64 MB of memory

Input: Keyboard and mouse

2.2 Recommended Requirements

OS: Windows XP (SP3)

CPU: 3.0 GHz or more

RAM: 2048 MB or more

Hard Drive: 20.0 GB or more free space

Video: Video Card with 256 MB of memory

Input: Keyboard and mouse

2.3 Tested Hard and Software

OS: Windows XP (SP1, SP2, SP3)

CPU:

- AMD XP 2400+ (2.0 GHz)
- AMD 64 3200+ (2.0 GHz)
- AMD 64 3800+ (2.4 GHz)
- Intel Centrino (1.5 GHz)

- Intel DualCore T3400 (2.16 GHz)
- Intel DualCore E6400 (2.2 GHz)
- Intel DualCore E8400 (3.0 GHz)

RAM: 512 MB - 2048 MB

Hard Drive: 10.0 GB or more free space

Video:

- ATI Radeon X800(256 MB)
- ATI - AMD Radeon HD4890 OC (1024 MB)
- Nvidia 4200 (64 MB)
- Nvidia FX 5900 ultra (256 MB)
- Nvidia 8800gt (512 MB)
- Intel GMA 4500 (shared memory)
- Intel 855GME (shared memory)

Input: Keyboard and mouse

The shared memory systems work fine without using the Combined View (3.5.8).

3 Program Overview

In the following few sections you get a detailed description of the whole application, its usage and appliance.

Primary the application was designed to calculate automatically the volume (area) of the heart of the zebrafish. To do this, you require a two dimensional video sequence with the pumping heart (which formats are supported will be explained later). Via the GUI you are able to resize and playback the whole sequence and perform various operations. Since it has not been possible to calculate the area of the heart automatically so far, the program provides you with many image manipulation and segmentation algorithms to calculate the area manually (step by step). Consequently the application can also be used to test various segmentation and motion tracking algorithms (especially from the OpenCV library (3.2)).

If the application is executed the first time you receive a blank screen and no sequence is loaded. This can be changed in the options menu to load the last recently used sequence automatically. After loading a sequence and calculating the correlation coefficient the application should look like in Figure 3.1.

3.1 General

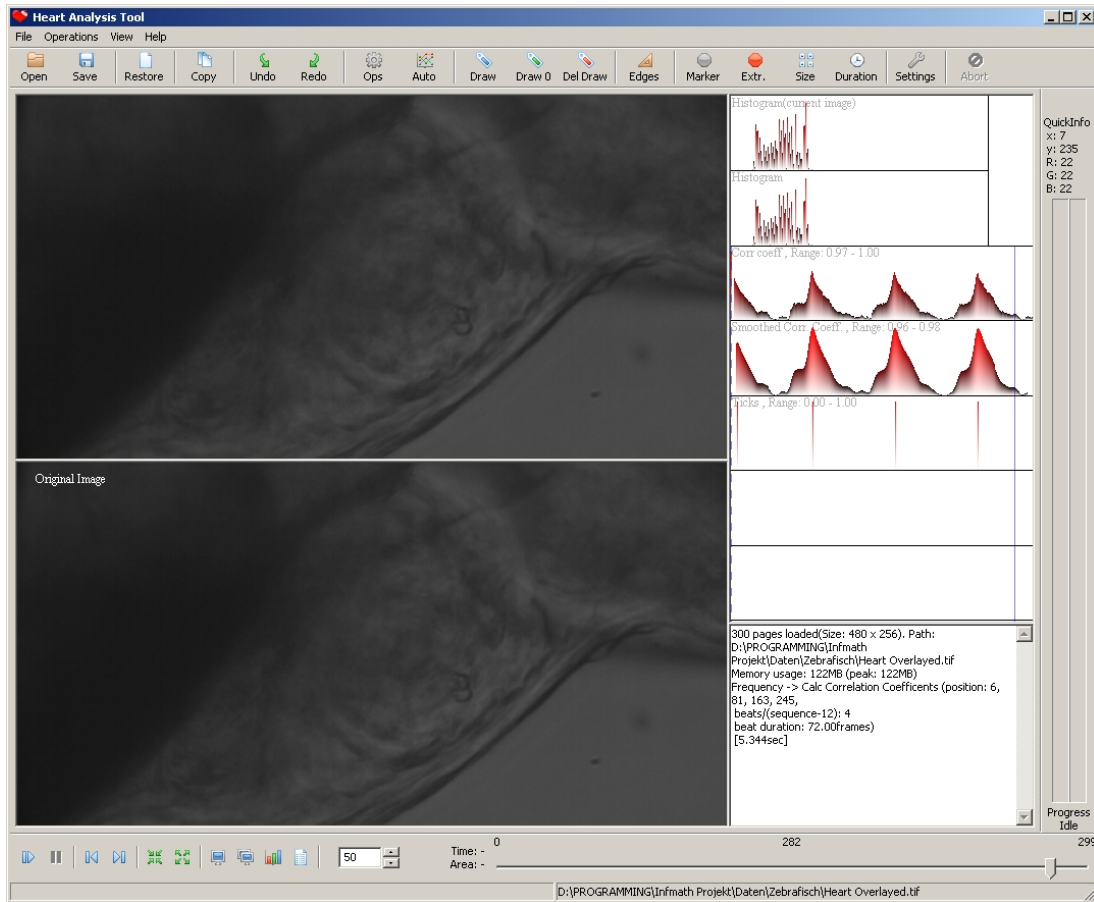
Some methods operate with a white background, others with a black background. Mathematically the background is black (0). The reason of either using a white or a black background is the better visibility of dark elements on a white background. The requirement of the foreground can be read at the specific operations at the *Operations* chapter. If you need another background for a specific method simply use Invert (4.5.1) or press the i button on the keyboard.

The application works only with one byte (unsigned char) grey scale images. As a result the range of integer values is from 0 to 255.

3.2 OpenCV

This application makes extensive use of the OpenCV library (Open Source Computer Vision library). It is a very fast and comprehensive image processing toolbox. As most methods of the OpenCV library are very complicated we suggest to read the OpenCV manual. This user manual doesn't describe most of those

3.3 Loading And Saving Sequences



methods. All OpenCV related operations are labelled with OpenCV in brackets. Download the OpenCV library including the user manual at: <http://sourceforge.net/projects/opencvlibrary/>

3.3 Loading And Saving Sequences

In the following section you can read all about how to save and open files and which formats are supported. It is suggested to use multi-page TIFF files. On the one hand you only have one big file, on the other hand the application is optimised to handle this format. Another possibility is the use of JPG, BMP, PNG or TIFF files. In that case the sequence is loaded from a set of images where you have to specify a source directory. The source directory should only contain the images for the sequence with the file extension as noted above to avoid errors. It is recommended to open at least a sequence with three images because most algorithms require three images. Further the application was not tested with

such a small sequence, even if it is possible to open a single image. Moreover the application tries to restore all primitives if a valid CSV file is available.

Same as loading you have the possibility to save the sequence into one big multi-page TIFF or as a set of JPG, BMP, PNG or TIFF files but again only the multi-page TIFF file is totally supported. Furthermore if you save the sequence as a multi-page TIFF, an additional CSV file is created. This CSV file contains all data except for the sequence itself. A detailed description of the structure of the CSV file follows in this section.

The following commands are listed in the file menu.

3.3.1 Open File

To open a file simply click on the first button of the Top Menu Bar (3.4) or select it via the top menu *File* → *Open File*. A file dialogue appears where you can choose the selected multi-page TIFF file. Remember that the file should contain at least three pages.

3.3.2 Open Directory

Use this command if you want to open a set of files from a directory. A dialogue appears where you can select your source directory.

Please read the top section to avoid problems.

3.3.3 Open File Resized

Choose this operation either on large sequences or to speed up the processing time. A dialogue appears where you can enter the new height of the image.

The new minimal height of the image have to be 64 pixel, otherwise it is set to 64 pixel automatically. If using images with a greater height than width it is possible that the resizing fails. Depending on the aspect ratio of your image you has to select a large size than 64 to receive a minimal width of 64 pixel. In case you receive a distorted image you should enable the Safe Mode (3.4.17). It is not suggested to set the new height greater than the original one as it does not cause better results.

This command only works on multi-page TIFF files.

3.3.4 Open Subsequence

Use this command on very large sequences (2000 pages and more with a resolution of 480x256) to select only a subset of all images. A dialogue appears, where you see the total number of images and where you can select the range of the subsequence.

This command works only on multi-page TIFF files.

3.3.5 Save File

Select this command to save the current sequence. An additional CSV file (section ref) with the same name will also be saved where you can find nearly all information like the values of the graphs or calculated velocity vectors. You can also execute this command via the second button of the Top Menu Bar (3.4).

3.3.6 Save Files

Choose this command if you want to save a set of images (files) instead of one multi-page TIFF file. If the dialogue appears, you can select the file format and the directory where the files are saved.

An additional CSV file will not be created.

3.3.7 Export Mask

If the required information is held in the mask you have the opportunity to save the mask as a multi-page TIFF file.

3.4 The Top Menu Bar

In the Top Menu Bar you find almost all important commands. If you highlight a button (keep the mouse a short time over the button without moving) a more detailed description appears. The menu is very intuitive so you may ignore this section. After loading a sequence The Top Menu Bar should look the same as in Figure 3.2. In case no sequence is loaded or the sequence is currently processed most buttons of the menu bar are disabled.

In the following sections the commands are listed as in the Top Menu Bar with the same name and the same order.

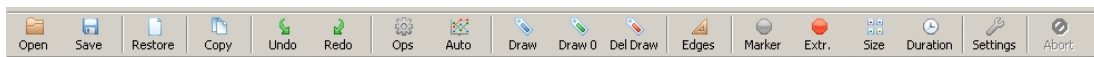


Figure 3.2: Top Menu Bar

3.4.1 Open

Identical to 3.3.1.

3.4.2 Save

Identical to 3.3.5.

3.4.3 Restore

On Restore the original sequence is reloaded and all graphic primitives like lines and arrows are deleted. As the original image is kept in memory, this command performs a lot of faster than reloading the sequence via Open File (3.3.1).

3.4.4 Copy

As in many other applications you can copy the current view (frame) to the clipboard. In case you have selected Combined, Single and Statistical View an image of the current sequence or graph is copied to the clipboard. After choosing the Log View the containing text is copied.

The image (frame) is copied to the clipboard as it is which means if you maximise the application you receive a higher resolution of the copied image. On specific resolutions it can happen that a distorted image is copied to the clipboard. In this case resizing the application in small steps helps.

3.4.5 Undo

On Undo the last operation is undone. The number of undoes depends on the number of buffered sequences. You can change this number in the Settings (3.4.17). Default you can restore one operation step.

Some data like the mask or midpoint can not be restored.

3.4.6 Redo

On Redo the last operation is redone. The number of redoes depends on the number of buffered sequences. You can change this number in the Settings (3.4.17). Default you can restore one operation step.

Some data like the mask or midpoint can not be restored.

3.4.7 Ops

This button opens a menu with all possible operations which can be applied to the sequence. As it is the most important and most extensive part it is described separately in chapter 4.

3.4.8 Auto

Identical to 4.1.

3.4.9 Draw (Drawing Mode)

Select Draw if you want to mark or surround a region in the sequence. You enter the drawing mode by clicking the Draw button. Then simply click into the current sequence to set the points of the polygon or spline (3.4.12). You abort drawing with a double click. Drawing is required for most Masking Operations (4.3).

It is suggested that your lines of your polygon do not overlap otherwise some operations may fail. Drawing can only be performed on the current sequence, neither in the Statistical View and nor in the Log View. On most operations only drawing at the first frame takes effect.

3.4.10 Draw 0

This mode is very similar to the previous one (3.4.9) but you can playback (3.5.1) the sequence while drawing; moreover the polygon or spline is only drawn on the first frame.

3.4.11 Del Draw

This command removes all graphic primitives of the current sequence.

3.4.12 Edges

This button allows you to toggle between polygons and splines (smooth curves). It is easier sometimes to start in polygon mode and switch to the spline mode to do the fine tuning afterwards.

3.4.13 Marker

For some operations (Export Line Profile, Size, Sonogram and Calc Luminance) you require a line as input. To draw a single line enter the marking mode via the Marker button and click first on the start position and afterwards on the end position. Accordingly you can select the preferred operation. Furthermore you can use the marker to calculate the size of a region. To receive the size in μm not in pixel you have to set the size before (3.4.15).

3.4.14 Extr

With this button you can manually set a frame number with the maximal extraction of the heart or segment. To do this select the frame with the slider of the Bottom Menu Bar 3.5 where the heart or segment seems to be largest and click the Extr button. A red line appears at your selected position in the Statistical View (3.5.9).

If no frame with the maximal extraction is selected the first one is taken.

3.4.15 Size

To calculate the real area or a distance a reference value is required. Use the Marker (3.4.13) to select a region from which you know the exact size. If you click on the Size button a dialogue appears where you can enter the size in μm . After doing this you receive the size in μm if using the marker. Furthermore the size of any calculated area is displayed in μm^2 (not in pixel).

Size specific values are displayed on the Statistical Window, on the Log Window or on the Bottom Menu Bar (3.5).

3.4.16 Duration

With this button you are able to set the real duration of the sequence in *ms*. After clicking the Duration button enter the real duration of the sequence in *ms*.

After assigning a duration to the sequence the elapsed time is displayed in the Bottom Menu Bar (3.5).

3.4.17 Settings

This button opens the Settings dialogue box which contains three tabs. Each tab will be described in the following three sections.

General Tab

In this tab you can set general options of the program.

- The first two labels displays the current default path for single and multi-page TIFF files. This path will be set, on a successful loading of a sequence. The path can not be changed.
- The next radio buttons give you the opportunity to start the application with the Single View (3.5.7) or Combined View (3.5.8). On slow systems or systems with an onboard video card it is suggested to start with the Single View. For more details read the Trouble Shooting (6).

To take effect you have to restart the application.

- If you set the check box *maximise on start* the application starts maximised. To take effect you have to restart the application.
- Default the program shows the required time of every operation in brackets in the Log Window. To disable the output uncheck the *show process time* box.

- The last checkbox gives you the opportunity to load the sequence automatically on start up.

This options only work on multi-page TIFF files.

Program Tab

Use this tab to configure program specific options.

To take effect you have to restart the application.

- In the first input field you can enter the buffer size. It defines the number of possible undoes and redoes. You receive the number of undoes and redoes by subtracting the input value with one. In case of two (default) you can undo and redo one operation once except for sequences. Use a value greater than two only on small sequences because the demand on memory grows enormously.
- Use start in save mode with an old video card which does not support power of two textures or OpenGL 2.0. It is also suggested to enable the safe mode if you work on small resolution sequences (smaller than 128x128 pixel).
- Depending on your system you can increase the performance by disabling the OpenGL text. If this checkbox is enabled you will not see any OpenGL texts as the minimum and maximum values of the Statistical Window. This option increases the refresh rate in the Combined- and Statistical View.
- The application uses smooth textures to render the sequence. This feature has the effect that the sequence can be stretched or extended and it is still looking good. If you disable it, each pixel is displayed. You can see the pixel as a square on zooming in.
- In the last checkbox you can decide whether you want to use a smooth blue or a normal silver progress bar. It is strongly recommended not to use the smooth one because it decreases the performance of the application enormously.

Preprocessing Tab

In this program you can choose between four operations which are executed automatically when loading a sequence.

- See Auto Normalise EX (4.5.4)
- See Auto Remove Flickering (4.5.3)
- See Auto Normalise (4.5.2)

- See Auto Correlation Coefficient (4.6.1)

It is useful to enable the Auto Correlation Coefficient checkbox.

3.4.18 Abort

This button is enabled if any operation is in process. If you abort the operation by clicking the button, the progress stops immediately. In most cases it is not possible to continue your work because the current sequence is distorted or incomplete. In this cases restore the sequence (3.4.3) or use the Undo button (3.4.5).

3.5 The Bottom Menu Bar

In the Bottom Menu Bar you can find most view specific controls. If you highlight a button (keep the mouse a short time over the button without moving) a more detailed description appears. The menu is very intuitive so you can ignore this section. After loading a sequence the bottom menu bar should look like as Figure 3.3. In the following sections the commands are listed like in the Top Menu Bar with the same name and the same order.



Figure 3.3: Bottom Menu Bar

3.5.1 Play

If you click on this button the playback of the sequence starts. You can set the playback speed with the Refresh Rate Control (3.5.11). Normally the playback is stopped if you start an operation, to decrease the progress time.

3.5.2 Pause

Click on the Pause button to stop the playback.

3.5.3 Display The Previous Frame

This button shows the previous frame.

3.5.4 Display The Next frame

This button shows the next Frame.

3.5.5 Original Size 1:1

If you want to display the sequence with the original aspect ratio and the original size use this button.

3.5.6 Stretch / Fit (Default)

This button fits the sequence into the window which looks best. You can also use this button after resizing or moving the sequence to restore the view. In some cases it is useful to have the correct aspect ratio or the original size. For this case use the button described before (3.5.5).

3.5.7 Single View

The Single View consists of one window where the current sequence is shown.

3.5.8 Combined View (Default)

The Combined View is a combination of all views. It consists of one window with the current sequence, another window with the original sequence, the Statistical Window and the Log. It is suggested to use this view because it is the most comfortable one and you see all necessary information simultaneously. On slow playback, disabling the OpenGL text may help.

3.5.9 Statistical View

The Statistical View consists of one window containing the histogram with some graphs like the correlation coefficient.

3.5.10 Log

This view logs all operations with their parameters for either reconstructing your current progress easily, or for reproducing the automatic calculations.

3.5.11 Refresh Rate Control

With this control you can adjust the refresh rate in *ms*. The range is between 20 *ms* and 2000 *ms*. To use refresh rates below 50 *ms* you require a fast system. On slow playback disable the OpenGL text (3.4.17).

3.5.12 Time Area Label

This label displays the time in *ms* and the area in μm^2 .

Before time and area are displayed you have to set the duration (3.4.16) and the area (3.4.15).

3.5.13 Position Slider

Use this control for sliding through the sequence or for going to a specific position.

3.6 The Side Bar



Figure 3.4: Side Bar

3.6.1 QuickInfo Label

This label gives you information about the current mouse position and the grey scale value at this position. In case of the Statistical View you get the value of the graph. The shown RGB value does not provide you the real value in case of masks or for example red lines. It only considers the colour information of the grey scale sequence.

3.6.2 Progress Bar

The left progress bar informs you about the overall progress whereas the right progress bar informs you about the temporary progress. On simple operations both bars work identically.

3.6.3 Progress Label

The progress label displays the overall process in percent same as the left progress bar.

3.7 Various Instructions

3.7.1 Zooming

You can zoom in and zoom out by simply scrolling the mouse wheel. You can reset the view by selecting Original Size 1:1 (3.5.5) or Stretch / Fit (3.5.6).

3.7.2 Moving

To move or drag the image or sequence hold the middle mouse button and move the mouse. The Statistical View can't be moved.

3.7.3 Moving Points

After Drawing Mode (3.4.9) you can move or change the polygon or spline by holding down the left mouse button on the yellow points and move it. You can stop moving the point by releasing the left mouse button. You are able to move all yellow points connected to red lines except the centre point.

3.7.4 Threshold

To perform a Binary Threshold (4.2.3) double click on the desired position into the histogram of the current frame (Statistical View).

3.7.5 Operations Menu

The most important menu can be called by clicking the right mouse button anywhere in the frame.

3.7.6 Status Bar

The left part of the status bar informs you about the memory usage or a more detailed description of the selected operation. The right part shows the current path of the sequence.

3.7.7 Export Line Profile

To export an intensity line profile select your desired region with the marker. Afterwards select Export Line Profile from the top menu (*File* → *Export* → *Export Line Profile*) and select the path to save the CSV file. The file contains all intensity values along a line of each frame. A shortened example of the file follows below.

```
Frame 0
120, 120, 124, 122, 118, 113, 113, 113, 117, ...
Frame 1
116, 114, 118, 113, 114, 109, 112, 113, 108, ...
Frame 2
121, 118, 120, 116, 113, 112, 118, 116, 120, ...
Frame 3
124, 122, 121, 117, 116, 114, 116, 108, 112, ...
Frame 4
125, 122, 124, 113, 113, 110, 112, 109, 113, ...
Frame 5
124, 121, 122, 114, 120, 116, 116, 112, 112, ...
Frame 6
120, 118, 120, 112, 114, 112, 113, 108, 109, ...
...
```

3.7.8 Toggle Mask

In some cases you want to see or use the mask but it is actually hidden. You can force to show or hide the mask by selecting *View* → *Toggle Mask* from the top menu.

3.7.9 The CSV File

On saving the file as a multi-page TIFF, a CSV file with all necessary information is created. An shortened example of the file follows below. The first few lines represent the header of the file and contain various information to validate the file. The proper data starts at the 12th line with *Duration [ms]*. Interpreting the file should be straightforward. First you have to locate the name of interest like *Duration [ms]*. After a line break the real value is arranged. In some cases either

a comment or a more detailed description is arranged between the name and the value like *Corr coeff*. $[S0]$ to $[S4]$ represent the values of the Statistical View. If the graph of the Statistical View is empty this section does not exist. $[I0]$ to $[I3]$ show the value of the graphic primitives like lines or arrows. For example if you want to evaluate the optical flow you require the arrow points of section $[I1]$. Pairs of values are separated by a comma, frames usually by a semicolon.

Heart Analysis Tool 1.0.0.0

Number of images
49

Dimension [width;height]
480;256

Path
D:\Daten\Zebrafisch\New 29.1.2008 50 Frames.csv

Duration [ms]
0

Size [um]
0

Size marker [pixel]
0

Marker pos [x1,y1;x2,y2]
0,0;0,0

Max extraction [frame number]
0

Center [x;y]
-1;-1

Information of the statistics view
[S0]
Corr coeff
1.000000;0.973490;0.973490;0.979962;

Internal data
[I0]
Points [frame number;x0,y0;x1,y1;...]

[I1]

Arrow points [frame number;x0,y0;x1,y1;...]

4 Operations

4.1 Auto

Automatic operations are a sequence of algorithms or operations with the advantage that you need no further knowledge of segmentation, motion tracking etcetera. To get an idea what the application causes simply open the sample sequence and perform the operations of this section. As it is only a set of algorithms you can read the details at the specific operations, only the name, the order and the overall result of the operations are mentioned. Used parameters are quoted in brackets.

4.1.1 Detect Area

This sequence detects the area of the heart without any pre knowledge or segmentation. It is very sensitive to noise which means that single error pixel influences the calculation of the area negatively.

Note: Use this method only for testing purposes.

- Image Enhancement → Remove Flickering
- Motion Tracking → Remove Background
- Thresholding → Threshold Auto [2]
- Smoothing → Median (OpenCV) [3]
- Masking → Convex Hull (OpenCV)

4.1.2 Find Centre

This method determines the centre by calculating the average of all defined motion pixel. Furthermore a calculation of the regression line is done to receive the orientation of the heart.

- Image Enhancement → Remove Flickering
- Motion Tracking → Remove Background
- Thresholding → Threshold Binary [247]

- Smoothing → Median(OpenCV) [3]
- Other → Find Center

4.1.3 Detect Area Ellipse

With this sequence you fit an ellipse around the heart. The size of the ellipses is determined by all counted motion pixel.

Use this method only for testing purposes.

- Image Enhancement → Remove Flickering
- Motion Tracking → Remove Background
- Smoothing → Gauss (OpenCV) [1.20]
- Thresholding → Threshold Binary [251]
- Other → Find Center
- Image Enhancement → Invert
- Other → Calculate Area
- Other → Calc Ellipses

4.1.4 Detect Area Motion

Similar to Detect Area (4.1.1) this method uses no pre knowledge or enhanced segmentation but in this case the motion pixel are determined by an OpenCV algorithm.

Use this method only for testing purposes.

- Image Enhancement → Remove Flickering
- Motion Tracking → Remove Background
- Image Enhancement → Normalise Image
- Segmentation → Segment Motion (OpenCV)
- Image Enhancement → Normalise Image
- Morphologic → Erode
- Thresholding → Threshold Binary [249]
- Image Enhancement → Invert
- Masking → Convex Hull (OpenCV)

4.1.5 Chan Vese 2D

This sequence detects the area of the heart by using the Chan Vese segmentation (stops after 20 iterations). It is very insensitive to noise and it produces smooth corners.

To get better results restrict the area with a mask at the first frame.

- Frequency → Calc Correlation Coefficients
- Image Enhancement → Remove Flickering
- Motion Tracking → Remove Background
- Thresholding → Threshold Auto [10]
- Image Enhancement → Invert
- Segmentation → Chan Vese [μ : 2.00; l1: 0.80; l2: 1.20; vis: 0; max it: 20]
- Segement → Detect Largest Segment
- Motion Tracking → Calculate Area
- Masking → Use As Mask

4.1.6 Chan Vese 2D Exact

It is the same sequence as Chan Vese 2D (4.1.5) but it stops either on saturation or after 1000 iterations.

4.1.7 Chan Vese 3D

This sequence detects the area of the heart by using the modified Chan Vese segmentation for three dimensional data (stops after 20 iterations). It is very insensitive to noise and it produces smooth corners. Chan Vese 3D yields to the best results.

To get better results restrict the area with a mask at the first frame.

- Frequency → Calc Correlation Coefficients
- Image Enhancement → Remove Flickering
- Motion Tracking → Remove Background
- Thresholding → Threshold Auto [10]
- Image Enhancement → Invert

- Segmentation → Chan Vese 3D [mu: 2.00; l1: 0.80; l2:1.20; max it: 20]
- Segement → Detect Largest Segment
- Motion Tracking → Calculate Area
- Masking → Use As Mask

4.1.8 Chan Vese 3D Exact

It is the same sequence as Chan Vese 3D (ref) but it stops either on saturation or after 1000 iterations.

4.1.9 Chan Vese 2D Ellipse

This sequence detects the area of the heart by using the Chan Vese segmentation (stops after 20 iterations). It is very insensitive to noise and it produces smooth corners. This method uses the data calculated by the Chan Vese algorithm to determine the size and position to fit an ellipse around the heart.

To get better results restrict the area with a mask at the first frame.

- Frequency → Calc Correlation Coefficients
- Image Enhancement → Remove Flickering
- Motion Tracking → Remove Background
- Thresholding → Threshold Auto [10]
- Image Enhancement → Invert
- Segmentation → Chan Vese [mu: 2.00; l1: 0.80; l2: 1.20; vis: 0; max it: 20]
- Segement → Detect Largest Segment
- Motion Tracking → Calculate Area
- Image Enhancement → Invert
- Other → Find Center
- Other → Calc Ellipses
- Masking → Use As Mask

4.2 Thresholding

Thresholding is very important for isolating certain pixel. In this section you get a brief introduction to various thresholding methods.

4.2.1 Threshold To White

All pixel with a value above the selected number are set to white all others remain constant.

4.2.2 Threshold To Black

All pixel with a value below the selected number are set to black all others remain constant.

4.2.3 Threshold Binary

All pixel with a value above the selected number are set to white all others to black. You can also execute the binary threshold by double clicking in the Statistical View into the histogram.

4.2.4 Threshold Auto

This method allows you to determine the value of the threshold for a given percentage and performs a Binary Threshold (4.2.3) with the caculated value. For example you estimate that 50% of your data is in range you enter 50 and recieve a threshold where approximate 50% of all pixel of the whole sequence are in range. Because of the usage of discrete values it may happen that you recieve a threshold of 58% or 46% instead of 50%. In the worst case you have to select another value since it is not possible to find a threshold with your chosen value.

4.2.5 Double Threshold

Use this threshold if you want to isolate a certain grey scale value. You are able to enter a lower and an upper threshold. All values between and equal are set to black, all others to white.

4.3 Masking

Masking is very important especially to obviate error pixel or regions which are not previously of interest. All necessary methods to perform this are described in the following section. A mask is displayed in a blue transparent colour. In most

cases you select the Drawing Mode (3.4.9) for defining the mask. Look out that the lines or curves of your region do not overlap.

4.3.1 Create Mask

First define a region with the Drawing Mode (3.4.9) and select Create Mask afterwards. The selected area (mask) is displayed in a blue transparent colour.

4.3.2 Cut Mask First Frame

If you want to cut a special region also called ROI (Region Of Interest) you only have to surround your preferred area at the first frame and execute this command. Consequently you only see your selected region, all other pixel are set to white.

4.3.3 Cut Mask All Frame

Similar to the previous method you can define a region of interest but in this case you have to draw a region at all frames.

4.3.4 Detect Background

This method uses different algorithms to determine the background or static elements. You can use this method followed by Cut Mask First Frame to automatic detect the region of interest.

4.3.5 Blend Out

It is sometimes useful to blend out the border. With this function you can define the width of the border in pixel for blending out. If you disable soft corners you only receive a expanded border without blending. You can also perform an inner and outer blending by disabling only outer mask.

4.3.6 Convex Hull (OpenCV)

As the name implies the function of the OpenCV library calculates the convex hull with the Slansky algorithm. All values smaller than 255 (white) are used for creating the hull.

4.3.7 Fit Line To Edges

Use this method if you have sharp (hard) edges. In that case draw a polygon near this corners and perform this operation afterwards to fit the polygon to the edges.

4.3.8 Trace Borders

Some operations return a mask as a result which is as storage intensive as the sequence itself. Trace Borders gives you the ability to draw a line stipple (contour) around the mask automatically.

4.3.9 Use As Mask

Use this command to set the current image as mask and restore the original sequence. All white pixel represent the mask.

4.4 Segmentation

In the following section you get a short introduction to some segmentation algorithm. In our tests the Chan Vese 3D algorithm have returned the best results.

4.4.1 Chan Vese 3D

This method performs a segmentation with the modified Chan Vese algorithm. As input use a binary image. The background has to be black (0) the foreground white (0) contrary to most other operations. To save time set a value below 100 for the maximum number of iterations. This method is very memory intensive so be careful you have enough free memory.

4.4.2 Chan Vese 2D Visualisation

It is similar to Chan Vese 2D (4.4.3) but in this case the actual progress is displayed in real time. This operation performs slower than without visualisation.

4.4.3 Chan Vese 2D

This method performs a segmentation with the Chan Vese algorithm. As an input use a binary image. The background has to be black (0) the foreground white (0) contrary to most other operations. To save time set a value below 100 for the maximum number of iterations.

4.4.4 Meanshift Segmentation (OpenCV)

For this segmentation use the original data so no preprocessing is necessary. To get further information read the OpenCV manual (3.2).

4.4.5 Pyramid Segmentation (OpenCV)

For this segmentation use the original data so no preprocessing is necessary. To get further information read the OpenCV manual (3.2).

4.4.6 Detect Largest Segment

This method detects the largest connected segment and closes its holes. It can also be used for closing holes only. The background has to be black (0) the foreground can be any colour except black.

4.4.7 Prepare Chan Vese

With this sequence you can prepare a binary image which is required for the Chan Vese segmentation.

4.5 Image Enhancement

In this section you find basic methods to improve the quality of the sequence (except for invert).

4.5.1 Invert

With this operation you can invert the image. Keep in mind that some methods require a black background others a white background especially when using binary images.

4.5.2 Normalise Image

Normalise Image scales (expands) the whole sequence between 0 and 255. Normally you gain a better contrast after using this method.

4.5.3 Remove Flickering

On some sequences you notice a jitter because the brightness changes as result of the illumination. Use this method to adjust the brightness automatically.

4.5.4 Normalise Image EX

Normalise Image EX scales each frame between 0 and 255. Since the scaling has been based on every single frame this method adjusts the brightness automatically.

4.5.5 Remove Flickering Extended

In this case the adjustment of the brightness is calculated from one of the four corners. On some sequences you have to use this method because Remove Flickering (4.5.3) does not lead to correct results. The corner for the regularisation is selected via the correlation coefficient.

4.6 Frequency

In the next section frequency related operations will be discussed.

4.6.1 Calc Correlation Coefficient

This method calculates the correlation between the first and all other frames. The result is displayed in the Statistical Window. Furthermore the frequency and pulse of the heart is calculated and displayed in the Log Window. If you want to compare the correlation coefficient with another except for the first frame select a different one with the max Extr button(3.4.14).

4.6.2 Calc Luminance

Before you can calculate the luminance you have to use the Marker. Select a region overlapping the heart. Calc Luminance determines the average brightness over all frames and displays the result in the Statistical Window.

4.6.3 Cross Correlation

Select the preferred area using the Drawing Mode. Similar to Calc Correlation Coefficient the Cross Correlation calculates the correlation between all frames but in this case only the ROI is used. An alternative is using Cut Mask All Frames to select your ROI and use Calc Correlation Coefficient afterwards. It should lead to more detailed results.

4.6.4 Sonogram

First you have to select a region overlapping the heart with the marker. If you select the Sonogram afterwards a small resizeable window with the sonogram is displayed.

4.7 Motion Tracking

In this section you find some algorithms like optical flow or segment motion to isolate the movement. The optical flow is displayed with arrows. Because of the different working of the three implementations you have to set the output size in pixel of the arrows. Consequently you have to possibilities; one is to take the average of the surrounded pixel the other is to display the optical flow of one pixel via the arrow. Further this means you only have the possibility to or.

4.7.1 Optical Flow (HS) (OpenCV)

This method performs a calculation of the optical flow using the Horn and Schunck algorithm. Read the OpenCV manual (3.2) for additional information.

4.7.2 Optical Flow (LK) (OpenCV)

This method performs a calculation of the optical flow using the Lucas Kanade algorithm. Read the OpenCV manual (3.2) for additional information.

4.7.3 Optical Flow (BM) (OpenCV)

This method performs a calculation of the optical flow using the block matching algorithm. Read the OpenCV manual (3.2) for additional information.

4.7.4 Segment Motion (OpenCV)

This method uses the motion tracking algorithm of the OpenCV library. It requires about 20 steps for initialisation. Read the OpenCV manual (3.2) for additional information.

4.7.5 Snake Image (OpenCV)

Snake Image works similar as Find Correlation. First you draw a polygon around the heart only at the first frame. The edge points should be set on significant positions like corners or other eye-catching spots. Notice that the lines itself are ignored, only the points are of interest. The algorithm tries to fit the polygon at all other frames.

4.7.6 Find Correlation

First you have to draw a polygon around the heart only at the first frame. The edge points should be set on significant positions like corners or other eye-catching spots. Notice that the lines itself are ignored, only the points are of interest. The

algorithm tries to trace the significant points using the correlation coefficient. You sometimes gain better results on an edge image.

4.7.7 Difference Image

Difference image simply subtracts the previous frame of the current one. The background is set to white for better visibility. We take the absolute values of the subtraction to get non-negative values. This method is very sensitive to noise.

4.7.8 Remove Background

This method subtracts the average image of all images. Normally the result is much better than the Difference Image variant. Same as in the previous method the background is set to white and absolute values are taken.

4.7.9 Remove Background (signed)

The only difference between to Remove Background and Remove Background (signed) is that no absolute values are taken. The background is set to grey (127), negative values are displayed darker, positive values brighter.

4.7.10 Remove Background EX

Unlike the previous versions the current state of the heart (decreasing or increasing) influences the result. For this you have to calculate the correlation coefficient before and set the maximum extraction. When the heart increases or decreases the previous and next frame is also used to determine movement pixel.

4.8 Smoothing

Smoothing is very important for processing images in general. In most cases the Gauss filter is sufficient.

4.8.1 Gauss (OpenCV)

Using the sigma value you can specify the intensity of smoothing. If you use a σ of 1.2 you receive a soft smoothing, if you use a σ of 4.0 you gain a very intensive smoothing.

4.8.2 Blur (OpenCV)

Read the OpenCV manual (3.2) for additional information.

4.8.3 Median (OpenCV)

The median filter is very effective against single error pixel.

4.8.4 Bilateral (OpenCV)

This filter reduces the colour space. Read the OpenCV manual (3.2) for additional information.

4.9 Edge Detection

In this section two edge detectors are described shortly.

4.9.1 Canny

This own implementation of the Canny edge detector has the advantage that it doesn't execute the thresholding with hysteresis for a better visibility of the strength of the corners.

4.9.2 Canny (OpenCV)

The Canny edge detector return good results for almost all tasks. It uses non-maximum-suppression to eliminate multiple corners and double thresholding with hysteresis to trace weak corners.

4.9.3 Sobel (OpenCV)

Use this detector only for primitive tasks because it only returns good results on absolute corners.

4.10 Morphologic

With the consecutive execution of dilation and erosion you can simulate the morphological closing and opening.

4.10.1 Dilate

Morphological dilation for grey scale images.

4.10.2 Erode

Morphological erosion for grey scale images.

4.11 Image Addition

This approach was implemented to find a connection between the single frame and the sum of some or all images.

4.11.1 Total Sum

This method calculates the average of all frames.

4.11.2 Total Sum Prepared

This method calculates the sum of all movement pixel and scales it.

4.11.3 Average Image

With this command you can sum a set of images and take the average of them. It has a similar effect as Gauss smoothing. Additionally the sequence seems to run slower and the movement is better visible.

4.12 Other

In this section you find some methods without a special classification.

4.12.1 Copy Image From

Depending on the settings responsible for the buffer you have the possibility to use a previous sequence instead of the current one.

4.12.2 Centre

This method calculates the centre of all non white pixel. It also calculates the regression line through a set of points.

4.12.3 Calculate Ellipses

If the area and the midpoint has already been calculated this method uses the information to fit an ellipse around the heart.

4.12.4 Calculate Area

If a mask is set this method calculates the number of all mask pixel, otherwise the number of all non white pixel.

5 Hotkeys

Hotkey	Command
A	Abort
B	Remove Background
C	Calc Correlation Coefficient
D	Draw
F	Remove Flickering
I	Invert
M	Create Mask
R	Restore
N	Normalise Image
S	Sobel
X	Set extraction
Z	Zero Draw
1 or Numpad 1	Detect Area
2 or Numpad 2	Find Center
3 or Numpad 3	Detect Area Ellipse
4 or Numpad 4	Detect Area Motion
5 or Numpad 5	Chan Vese 2D
6 or Numpad 6	Chan Vese 2D Exact
7 or Numpad 7	Chan Vese 3D
8 or Numpad 8	Chan Vese 3D Exact
9 or Numpad 9	Chan Vese 2D + Ellipse
F1	Help
F3	Stretch
F4	1:1
F5	Single view
F6	Combined view
F7	Stat view
F8	Log
Space	Toggle play pause
Num +	Strech
Num -	1:1
Alt + F4	Exit

Hotkey	Command
Alt + S	Settings
Strg + A	Abort
Strg + C	Copy
Strg + D	Draw
Strg + S	Draw Zero
Strg + M	Marker
Strg + O	Open
Strg + R	Restore
Strg + S	Save
Strg + X	Exit
Strg + Y	Redo
Strg + Z	Undo
Left Cursor	Previous image
Right Cursor	Next image
Entf	Del Draw

Note: On some keyboards Strg is labelled as Ctrl and Entf as Del.

6 Trouble Shooting

In some cases systems with a video card with shared memory have problems to show the Combined View correctly. In this case open the program settings and select Single View on the first page (General tab). To take effect you have to restart the program. After restarting it, the program displays the Single View instead of the Combined View (default). If you close the application it might happen that an error message appears which can be ignored. It is recommended not to use the Combined View in this situation.

Another problem can appear on systems with an old video card which doesn't support non power of two textures. In this case an error may be shown in the Log Window and the sequence will not be shown correctly or the window will may be kept blank. Simply open the program settings and select on the second page (Program tab) start in safe mode.

One major problem is the huge amount of memory usage of the program. If the opened TIFF sequence is near on the limit of free memory, it can lead to program termination. In most cases you will receive an error message in the Log Window, if you try to open the sequence, which informs you, that the sequence exceeds your free memory. In that case you have two possibilities:

1. Open the sequence resized.
2. Open only a subsequence.

You can find both commands in the file menu. In particular have a look at the Chan Vese 3D algorithm. If the program crashes at the beginning of the operation try to reopen the sequence with the two suggestions mentioned before.

The application writes some settings to the Windows registry. If any tool is blocking the access to the registry, please add the *Heart Analysis Tool* to the exceptions. If you want to delete the registry entry go to *Start* → *Run* and type `regedit` into the prompt and delete the entry `HKEY_CURRENT_USER` → *Software* → *Heart Analysis Tool*.

It might happen that you get two versions of the program:

1. Heart Analysis Tool.exe
2. Heart Analysis Tool(colour).exe

The second version supports the colour output. This feature visualises contents with colour and it is currently in experimental state. The colour visualisation was

removed from the first version because it takes more memory which is strongly required to open large sequences. It is recommended to use the first version.

Whenever you have a question feel free to contact me.
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